1.5 Autoimmunity and the endocrine system

Introduction

The immune system plays a vital role in protecting the body against attack from bacteria, viruses, and other foreign bodies. Essential to this function is its ability to distinguish between self and non-self. Breakdown in this ability can occur, causing an inappropriate immune response against self-tissues and organs, known as autoimmunity. Endocrine autoimmunity is characterized by autoantibody production against components of the endocrine system (Table 1.4), resulting in a variety of conditions including, for example, type 1 diabetes, Graves' disease, Hashimoto's thyroiditis, Addison's disease, and autoimmune polyendocrine syndrome type 1. Although, the exact organ/s targeted for autoimmune attack vary from disease to disease, co-clustering of different endocrine diseases within individuals and families, and the identification of shared immunological genetic susceptibility loci between diseases, suggest the presence of common pathogenic pathways. This section will focus on how disruption in immunological pathways contributes to disease onset.

Breakdown in ability to distinguish self from non-self

Constant monitoring of antigens is performed by T lymphocytes and is essential to ensure that foreign antigens are recognized quickly and an immune response mounted. A T cell's ability to distinguish self from non-self is key to this process and is achieved through central tolerance.

Central tolerance

CD4+ T helper (Th) cells, which activate B cells to produce antibodies, and CD8+ T cells, which produce cytotoxic T cells and natural killer (NK) cells, start off as common progenitor CD4−/CD8− T cells within the bone marrow. Progenitor CD4−/CD8− T cells undergo random rearrangement of their T-cell receptor (TCR) gene, creating precursor CD4+/CD8+ T cells. Random TCR rearrangements provide a mature T-cell repertoire capable of binding a vast array of antigens. Inevitably, however, some non-functional and self-/autoreactive precursor T cells are generated. Before entering the peripheral immune system, precursor T cells move to the thymus to undergo selection.

Positive selection

Precursor CD4+/CD8+ T cells' TCR functionality is tested by determining binding to human leukocyte antigen (HLA) molecules presenting antigens. Precursor T cells that bind antigen-presenting HLA molecules receive a survival signal, whereas TCRs that do not bind receive no survival signal and so die.

Table 1.4 Different autoantigens identified in the endocrine autoimmune diseases

Breakdown in ability to distinguish self from non-self
Disruption in antigen presentation to T cells
Alterations in T-cell activation/signalling
Variation in B-cell function
Environmental impact on the immune system

Adapted from Wass JAH, Stewart PM, Amiel SA. et al., *Oxford Textbook of Endocrinology and Diabetes*, 2nd edition, Table 1.6.2, pp. 34–44 Copyright © 2011, with permission from Oxford University Press

Negative selection

Precursor T cells that survive positive selection are then retested to determine if they bind HLA molecules presenting an array of self-antigens. Any T cells that recognize self-antigens too strongly, suggesting autoreactivity, undergo TCR editing to alter antigen specificity or undergo apoptosis. Under normal circumstances, only functional, non-autoreactive CD4+/CD8+ precursor T cells mature into CD4+ Th cells or CD8+ T cells and are released into the periphery.

Autoimmunity: A case of bad education?

Many endocrine antigens are expressed outside of the thymus, raising the question as to whether precursor T cells have been educated to recognize them. The protein encoded by *AIRE*, defects in which were originally detected as the genetic cause of autoimmune polyendocrine syndrome type 1, was found to transcribe otherwise tissue-restricted antigens in the thymus, including several thyroid and pancreas antigens. Support for a role of disrupted central tolerance in autoimmune disease came from screening a variable number of tandem repeats upstream of the insulin gene in type 1 diabetes. The region consists of 14–15 base pairs of consensus sequence clustered into Class I (30–60 repeats), Class II (60–120 repeats), and Class III (120–170 repeats) alleles. The presence of Class I alleles predisposes for type 1 diabetes, whereas presence of Class III alleles protects against type 1 diabetes. Functional studies demonstrated that Class III alleles encode 200%–300% higher insulin transcription in the thymus, compared to Class I alleles, suggesting that variation in the thymic transcript levels of endocrine genes could affect how successfully negative selection removes autoreactive T cells.

Disruption in antigen presentation to T cells

The constant processing and display of antigens by HLA molecules on the surface of antigen-presenting cells (APCs) in the periphery is essential for T-cell screening and, if appropriate, the generation of an immune response (see Figure 1.5). Processing of endogenous (internal) and exogenous (external) antigens is achieved through two distinct pathways. Endogenously derived proteins, including viral proteins, are initially ubiquitinated and then degraded by the cytosolic pathway. This leads to the generation of peptides which enter the endoplasmic reticulum, where they become bound by HLA class I molecules. Antigen-bound HLA class I molecules exit the endoplasmic reticulum and translocate to the APC surface for presentation to CD8+ T cells. If the antigen is recognized as non-self, the CD8+ T cell becomes activated, leading to generation of cytotoxic T cells, which destroy infected cells, and NK cell activation, which produce lymphokines, cytokines, and chemokines essential for immune cell recruitment and cell destruction. Exogenous proteins, such as those from bacteria, have to be internalized into the APC. Once internalized,

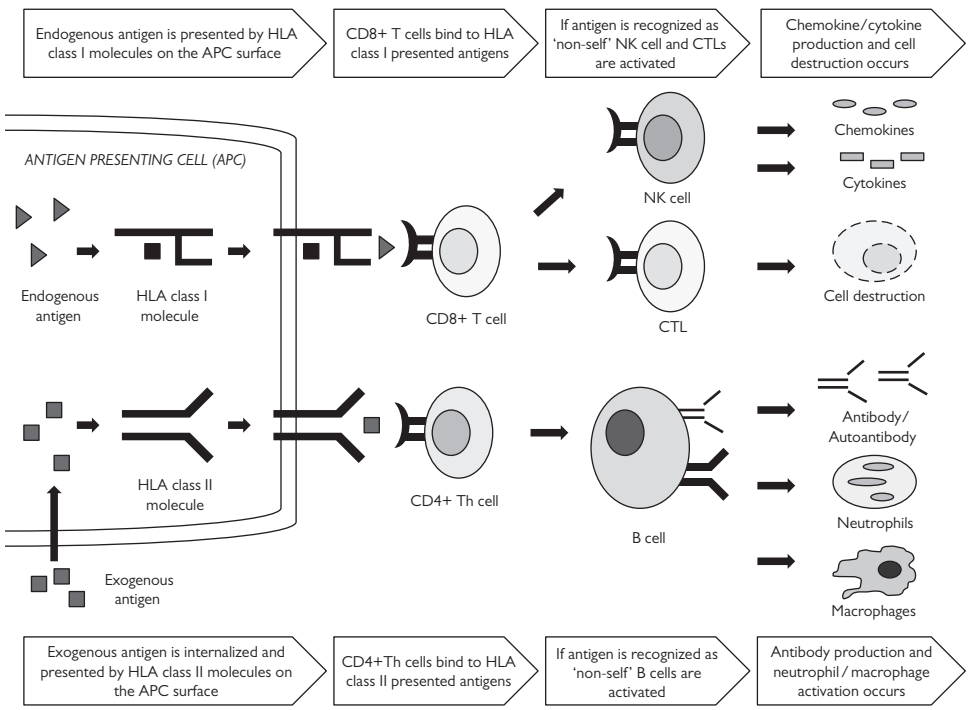

Fig 1.5 The role of T and B cells in monitoring antigens and mounting an immune response against any non-self antigen; HLA, human leukocyte antigen; Th, T helper; NK, natural killer, CTL, cytotoxic T lymphocyte.

exogenous proteins navigate between a series of increasingly acidic compartments, known as the endocytic pathway, leading to the generation of antigens. These antigens are bound by HLA class II molecules, which exit the endocytic pathway and are displayed on the APC surface for recognition by CD4+ Th cells. If the antigen is recognized as non-self, B-cell activation/antibody production and macrophage activation occurs. The vital role of the HLA region in the immunological defence system is only possible because of the highly variable nature of its encoding genes, which enable the human population to encounter and survive such a wide array of antigenic threats. Some of these variants, however, can predispose to the autoimmune disease process.

Variation in exogenous antigen presentation by HLA class II molecules

Variation within the HLA class II region-encoded HLA-DR molecule (composed of a HLA-DRB1 chain and the almost non-polymorphic HLA-DRA1 chain) and the HLA-DQ molecule (composed of a HLA-DQB1 chain and a HLA-DQA1 chain) has been examined, with the DRB1*03 variant being strongly associated with Graves' disease and type 1 diabetes, and DRB1*04 being strongly associated with type 1 diabetes and Hashimoto's thyroiditis. Due to linkage disequilibrium among DRB1, DQB1, and DQA1 (see Chapter 4), the haplotypes containing DRB1*03 and DRB1*04, known as the DR3 and DR4 haplotypes, respectively, are also strongly associated with autoimmune disease. Regression analysis of DRB1–DQB1–DQA1 haplotypes in Graves' disease revealed the association was due exclusively to DRB1 and DQA1. When comparing the DRB1*03 molecule, which predisposes to Graves' disease, to the DRB1*07 molecule, which protects against Graves' disease, change at amino acid position 74, from a positively charged arginine to a non-charged glutamine, respectively, was observed, in an area which forms part of the HLA antigen-binding domain. Variation at position 74 also differentiated DRB1*0403 and DRB1*0406 molecules, which contain negatively charged glutamic acid at that position and confer a low risk for type 1 diabetes, from high-risk alleles, which contain non-charged polar amino acids at that position. Several hypotheses have been suggested to explain how variation in antigen-binding pockets of HLA class II molecules could lead to autoimmune onset. These include:

- variation in the antigen-binding domain may cause preferential selection of a limited set of self-peptides during negative selection, allowing more autoreactive T cells to escape central tolerance
- preferential binding by a given DR/DQ molecule in heterozygous individuals may lead to presentation of specific antigens, depending on whether it is predisposing, neutral, or protective
- although HLA class II molecules present exogenous antigens, cross-presentation of endogenous antigens (normally presented by HLA class I molecules) can

occur, and vice versa, potentially altering how exogenous antigens are recognized by the immune system

Variation in endogenous antigen presentation by HLA class I molecules

Association of HLA class I molecules, such as HLA-A, HLA-B, and HLA-C, with many autoimmune endocrine conditions has been detected, with several hypothesis attempting to explain how they could be linked to autoimmunity:

- viral antigens preferentially presented by certain HLA class I molecules may be similar to self-antigens; an immune response mounted against these antigens could unintentionally cross-react with similar self-antigens
- viruses presented by HLA class I molecules can trigger strong immune responses that could unintentionally cross-react with self-antigens
- HLA class I molecules interact with killer-cell immunoglobulin-like receptors which help control NK cell activation, and variation in these interactions could cause inappropriate NK cell activation
- variation in HLA class I molecules could cause alterations in central tolerance mechanisms and affect T-cell education and/or the production of T regulatory cells

Variation in T-cell activation/signalling

For T-cell activation to occur, this requires not only binding of the TCR to the antigen presented by HLA molecules on the surface of APCs, but also co-stimulatory signals. These signals come from accessory molecules such as CD28, which promotes signalling, and CTLA-4, which downregulates signalling. CTLA-4 is upregulated during T-cell signalling and has been proposed to block signalling through numerous pathways. These include the following:

- binding of CD28 to co-stimulatory molecules CD80/CD86 can be blocked by CTLA-4
- CTLA-4 can alter lipid raft and microcluster formation, thus aiding downstream TCR signal transduction
- co-factors required to enable the TCR to bind to antigens presented by HLA molecules and determine if the antigen is self or non-self are controlled by CTLA-4
- CTLA-4 also produces negative signals to prevent T-cell activation
- CTLA-4 can alter the stability and strength of TCR and APC interactions by decreasing adhesion molecules required for such interactions

Genetic variants within CTLA-4 have been associated with multiple autoimmune disorders, including type 1 diabetes and Graves' disease (see also Chapter 2, Section 2.4) and have been shown to alter the ratio of full-length CTLA-4 to a soluble isoform. As it has been suggested that the soluble isoform may provide greater inhibition of T-cell activation than the full-length isoform, changes in the expression of the soluble isoform could alter T-cell activation thresholds. Other inhibitors of T-cell activation have also been associated with endocrine autoimmunity. Variation within *PTPN22* has been shown to encode a change from arginine to tryptophan at amino acid position 620 (referred to as R620W) of the LYP molecule encoded by *PTPN22*. The presence of the LYP620W variant has been shown to impair LYP interaction with Csk, an important intracellular inhibitor of the T-cell signal transducer Lck, causing greater inhibition of T-cell signalling. The presence of LYP620W could lead to stronger downstream inhibition of T-cell signalling, potentially altering autoreactive T-cell activity during central tolerance and thus affecting negative selection.

Although central tolerance removes the majority of autoreactive T cells before they enter the periphery, some autoreactive T cells do enter the periphery. T regulatory cells (a form of CD4+ Th cells expressing high CD25 and foxp3 levels) account for 6%–7% of the mature CD4+ Th cell population and suppress the activation and expansion of autoreactive T cells in the periphery. Not only could variation in CTLA-4 and *PTPN22* affect the function of T regulatory cells but variation in CD25 levels has been associated with Graves' disease and type 1 diabetes. This leads to alterations in interleukin 2 binding (CD25 forms part of the interleukin 2 receptor), which is involved in the development of T regulatory cells and their ability to cause autoreactive T cells to undergo apoptosis. With evidence emerging for a role for foxp3 variation in autoimmune disease onset, taken together this suggests alterations in policing of peripheral T-cell autoreactivity could contribute to autoimmunity.

Variation in B-cell function

For many years it has been postulated that B cells are merely involved in initiating autoimmunity by producing antibodies, whereas T cells progress disease. Increases in our knowledge of B-cell function has identified additional roles in the immune system, including acting as APCs to CD4+ Th cells in low-antigen environments, controlling inflammation through cytokine production, and being directly activated by Toll-like receptor ligands, such as bacterial DNA, independently of T cells and through the possible existence of B regulatory cells. A role for variation in molecules involved in B-cell signalling, such as B-cell activating factor and Fc receptor-like molecules, has been identified in disease, suggesting that the role of B cells in autoimmunity may be more complex than first envisaged.

Environmental impact on the immune system

It has been estimated that environmental factors make up >20% of the contribution to endocrine disease, and several of those identified, including viral and bacterial infection, increased hygiene, stress, smoking, and early intake of cow's milk, have been proposed to alter immune function (Table 1.5).

Manipulating our understanding of immune disruption to create improved therapeutics

Insights into the immune dysfunction behind autoimmune endocrine disease not only provide greater understanding of disease onset and progression but also identify immune pathways upon which therapeutic intervention could be undertaken. Whilst still in its infancy, immune modulation in autoimmune disease pathways will undoubtedly in the future provide improved therapeutic options for these diseases.

Table 1.5 Proposed environmental impacts on autoimmune endocrine disease and how they are believed to impact on the immune system

Environmental factor	How environmental factor is proposed to alter immune system
Viruses and bacteria	Birth rate data from type 1 diabetes and Graves' disease patients show peaks in autumn and winter, when viruses and bacteria are more virulent, compared to birth rates peaks in the summer and spring in the general population; however, more recently, work in several large Caucasian Graves' disease collections failed to show a peak in birth rates in summer and spring, casting doubt over the role of month of birth effects on autoimmune disease onset
Improved hygiene	Humans have developed strong, highly effective immune systems to enable them to survive any foreign threat encountered; due to increases in hygiene, coupled with changes in social behaviour, the body is encountering less foreign insults and, as a result, our highly primed immune systems could turn against us, causing autoimmune onset
Stress and smoking	These have immunosuppressive effects on the immune system by stimulating the hypothalamic–pituitary–adrenal axis, downregulating immune responsiveness and regulation
Exposure to cow's milk early in life	Exposure to cow's milk early in childhood, rather than breast milk, which provides immune support, has been proposed to contribute to type 1 diabetes, as the immune systems could trigger a response against cow insulin that may cross-react with human insulin; however, to date, no clear evidence of a role for cow's milk in increasing rates of type 1 diabetes has been established

Further reading

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